L3 ANSWER 1 OF 15 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI New human ena/**VASP**-like protein splice variant (EVL1), useful for preparing a composition for diagnosing or treating a disease or condition associated with decreased expression or overexpression of functional EVL1 e.g., cancer.
IN CORLEY, N C; GUEGLER, K J; LAL, P
AB US2004013670 A UPAB: 20040218
NOVELTY - A new isolated polypeptide comprises:
(1) a sequence having 418 amino acids, or its biologically active or immunogenic fragment; or
(2) a naturally occurring amino acid sequence having at least 95% identity with (1).
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:
(1) an isolated polynucleotide encoding the polypeptide;
(2) a recombinant polynucleotide comprising a promoter sequence operably linked to the polynucleotide;
(3) a cell transformed with the recombinant polynucleotide;
(4) a transgenic organism comprising the recombinant polynucleotide;
(5) a method of producing the polypeptide;
(6) an isolated antibody that specifically binds to the polypeptide;
(7) a method of detecting a target polynucleotide in a sample;
(8) a composition;
(9) a method for treating a disease or condition associated with decreased expression or overexpression of functional human ena/**VASP**-like protein splice variant (EVL1);
(10) a method of screening a compound for effectiveness as an agonist or antagonist of the polypeptide or in altering expression of the target polynucleotide;
(11) a method of screening a compound that specifically binds to, or that modulates the activity of, the polypeptide;
(12) a method of assessing toxicity of a test compound;
(13) a diagnostic test for a condition or disease associated with the expression of EVL1 in a biological sample;
(14) a method of diagnosing a condition or disease associated with the expression of EVL1 in a subject;
(15) a method of preparing a polyclonal or **monoclonal** antibody;
(16) a method of detecting the polypeptide in a sample;
(17) a method of purifying the polypeptide;
(18) a method of generating an expression profile of a sample that contains polynucleotides; and
(19) an array comprising different nucleotide molecules affixed in distinct physical locations on a solid substrate, where at least one of the nucleotide molecules comprises a first oligonucleotide or polynucleotide sequence specifically hybridizable with at least 30 contiguous nucleotides of the target polynucleotide.
ACTIVITY - Cytostatic; Anorectic; Immunosuppressive.
No biological data given.

MECHANISM OF ACTION - Gene therapy; EVL1-Antagonist; EVL1-Agonist. No biological data given.

USE - The polypeptide is useful for preparing a composition for diagnosing or treating a disease or condition associated with decreased expression or overexpression of functional EVL1 (claimed) e.g., autoimmune disorders, obesity or cancer.

Dwg.0/3

L3 ANSWER 2 OF 15 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 1
TI Method for the screening of chemical compounds for modulation of the interaction between an EVH1 domain or a protein with an EVH1 domain and an EVH1 binding domain or a protein with an EVH1 binding domain, and a method for detection of interaction
IN Jordan, Birgit; Druckes, Peter; Jarchau, Thomas; Walter, Ulrich
SO PCT Int. Appl., 34 pp.
CODEN: PIXXD2
AB The invention discloses methods for the identification of chemical compds. which can modulate the interaction between **VASP** and zyxin. **VASP** or a **VASP** derivative and zyxin or a zyxin derivative, which interact with each other, are brought into contact with a chemical compound under investigation. The influence on the interaction can be determined by means of antibodies for **VASP** or a **VASP** derivative and/or zyxin or a zyxin derivative, or by means of fluorescent markers on **VASP** or a **VASP** derivative and zyxin or a zyxin derivative

L3 ANSWER 3 OF 15 CAPLUS COPYRIGHT 2004 ACS on STN
TI Endocrine disruptor screening using DNA chips of endocrine disruptor-responsive genes
IN Kondo, Akihiro; Takeda, Takeshi; Mizutani, Shigetoshi; Tsujimoto, Yoshimasa; Takashima, Ryokichi; Enoki, Yuki; Kato, Ikunoshin
SO Jpn. Kokai Tokkyo Koho, 386 pp.
CODEN: JKXXAF
AB A method and kit for detecting endocrine-disrupting chems. using DNA microarrays are claimed. The method comprises preparing a nucleic acid sample containing mRNAs or cDNAs originating in cells, tissues, or organisms which have been brought into contact with a sample containing the endocrine disruptor. The nucleic acid sample is hybridized with DNA microarrays having genes affected by the endocrine disruptor or DNA fragments originating in these genes have been fixed. The results obtained are then compared with the results obtained with the control sample to select the gene affected by the endocrine disruptor. Genes whose expression is altered by tri-Bu tin, 4-octaphenol, 4-nonylphenol, di-N-Bu phthalate, dichlorohexyl phthalate, octachlorostyrene, benzophenone, diethylhexyl phthalate, diethylstilbestrol (DES), and 17-β estradiol (E2), were

found in mice by DNA chip anal.

L3 ANSWER 4 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 2

TI Zyxin is not colocalized with **vasodilator-stimulated phosphoprotein** (**VASP**) at lamellipodial tips and exhibits different dynamics to vinculin, paxillin, and **VASP** in focal adhesions.

AU Rottner, Klemens; Krause, Matthias; Gimona, Mario; Small, J. Victor; Wehland, Juergen [Reprint author]

SO Molecular Biology of the Cell, (October, 2001) Vol. 12, No. 10, pp. 3103-3113. print.
CODEN: MBCEEV. ISSN: 1059-1524.

AB Actin polymerization is accompanied by the formation of protein complexes that link extracellular signals to sites of actin assembly such as membrane ruffles and focal adhesions. One candidate recently implicated in these processes is the LIM domain protein zyxin, which can bind both Ena/**vasodilator-stimulated phosphoprotein** (**VASP**) proteins and the actin filament cross-linking protein alpha-actinin. To characterize the localization and dynamics of zyxin in detail, we generated both **monoclonal** antibodies and a green fluorescent protein (GFP)-fusion construct. The antibodies colocalized with ectopically expressed GFP-**VASP** at focal adhesions and along stress fibers, but failed to label lamellipodial and filopodial tips, which also recruit Ena/**VASP** proteins. Likewise, neither microinjected, fluorescently labeled zyxin antibodies nor ectopically expressed GFP-zyxin were recruited to these latter sites in live cells, whereas both probes incorporated into focal adhesions and stress fibers. Comparing the dynamics of zyxin with that of the focal adhesion protein vinculin revealed that both proteins incorporated simultaneously into newly formed adhesions. However, during spontaneous or induced focal adhesion disassembly, zyxin delocalization preceded that of either vinculin or paxillin. Together, these data identify zyxin as an early target for signals leading to adhesion disassembly, but exclude its role in recruiting Ena/**VASP** proteins to the tips of lamellipodia and filopodia.

L3 ANSWER 5 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

TI Endocannibinoid 2-arachidonoyl glycerol modulates ET-1-induced brain endothelium responses.

AU Spatz, M. [Reprint author]; Chen, Y. [Reprint author]; Golech, S.; Bembry, J. [Reprint author]; Mechoulam, R.; McCarron, R. M.

SO Society for Neuroscience Abstracts, (2001) Vol. 27, No. 2, pp. 2129. print.
Meeting Info.: 31st Annual Meeting of the Society for Neuroscience. San Diego, California, USA. November 10-15, 2001.
ISSN: 0190-5295.

AB Cerebral vascular endothelial cells (EC) play an active role in regulating vascular tone, blood flow and blood-brain barrier function. Previous experiments implicate circulating and locally produced vasoactive

mediators such as nitric oxide (NO) and endothelin-1 (ET-1). The endocannabinoid 2-arachidonoyl glycerol (2-AG) represents a novel vasoactive modulator that counteracts endothelial responses to ET-1. Ca2+ accumulation was assessed in human EC with fluo-3/AM. EC pretreated with buffer or 2-AG (5 or 15 min) were exposed to 20 nM ET-1 (30 sec); the resulting changes in cytoskeleton F-actin and vimentin were evaluated by staining with Texas Red-x phalloidin and **monoclonal** anti-vimentin antibody, respectively. **Vasodilator-stimulated phosphoprotein** (**VASP**) was assessed by Western blot analysis. The findings demonstrate that 2-AG counteracts Ca2+ mobilization and cytoskeleton rearrangement induced by ET-1. This event is mediated by the cannabimimetic CB1-receptor, G-protein, phosphoinositol signal transduction pathway, high (KCl), and Ca2+-activated K+ channels and were independent of NO, cyclooxygenase and lipoxygenase. The 2-AG-mediated effects on cytoskeleton and **VASP** (phosphorylation) involved PKC and cAMP, repsectively. These observations indicate that cannabinoid CB1-receptors play a role in EC function and indicate the potential existence of an alternative pathway to NO for abrogating ET-1-induced effects in the brain. This may be particularly relevant when endothelial responses to NO are down-regulated (i.e., hypertension, arteriosclerosis, hemorrhagic shock and Diabetes mellitus).

L3 ANSWER 6 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 3

TI A cGMP-dependent protein kinase assay for high throughput screening based on time-resolved fluorescence resonance energy transfer.

AU Bader, Benjamin; Butt, Elke; Palmetshofer, Alois; Walter, Ulrich [Reprint author]; Jarchau, Thomas; Drueckes, Peter

SO Journal of Biomolecular Screening, (August, 2001) Vol. 6, No. 4, pp. 255-264. print.
ISSN: 1087-0571.

AB Activation of cyclic GMP-dependent protein kinase (cGK) is an important event in the regulation of blood pressure and platelet function. Upstream signals are the generation of nitric oxide (NO) by NO synthases and the subsequent rise in cyclic GMP levels mediated by NO-dependent guanylyl cyclases (GCs). The identification of new cGK activators by high throughput sreening (HTS) may lead to the development of a novel class of therapeutics for the treatment of cardiovascular diseases. Therefore, a homogeneous, nonradioactive assay for cGK activity was developed using a biotinylated peptide derived from **vasodilator-stimulated phosphoprotein** (**VASP**), a well-characterized natural cGK substrate. The phosphorylated peptide could be detected by a **VASP** -specific **monoclonal** phosphoserine antibody and a fluorescent detection system consisting of a europium-labeled secondary antibody and allophyrocyanin (APC)-labeled streptavidin. Fluorescence resonance energy transfer (FRET) from europium to APC was detected in a time-resolved fashion (TR-FRET). Activation and inhibition constants for known substances determined by this new fluorescence-based assay correlated well

with published results obtained by conventional radioactive cGK activity assays. The assay proved to be sensitive, robust, highly specific for cGK, and suitable for HTS in 96- and 384-well formats. This assay is applicable to purified enzymes as well as to complex samples such as human platelet extracts.

L3 ANSWER 7 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 4

TI Age-related decrease of protein kinase G activation in vascular smooth muscle cells.

AU Lin, Ching-Shwun [Reprint author]; Liu, Xiaowei; Tu, Richard; Chow, Sylvia; Lue, Tom F.

SO Biochemical and Biophysical Research Communications, (September 14, 2001) Vol. 287, No. 1, pp. 244-248. print.
CODEN: BBRCA9. ISSN: 0006-291X.

AB Protein kinase G-I (PKG-I) activation is essential for vascular relaxation; however, its quantitative analysis in intact cells has been difficult. To overcome this difficulty, a **monoclonal** antibody, **VASP**-16C2, was recently developed that detects phosphorylated serine residue 239 of **vasodilator-stimulated phosphoprotein** (**VASP**), a substrate of PKG-I. In this study, we used this antibody to examine (i) possible functional differences between the alpha and beta isoforms of PKG-I, (ii) ability of cAMP to activate PKG-I, as compared to cGMP, the principal PKG-I-activating cyclic nucleotide, and (iii) time course and levels of PKG-I activation in vascular smooth muscle cells (VSMC) of young and old rats. We created COS-7 cell clones that overexpressed PKG-Ialpha or PKG-Ibeta, treated them with cAMP or cGMP, and analyzed their cell lysates for reactivity with **VASP**-16C2. The results showed that PKG-Ialpha phosphorylated **VASP** at a higher level than PKG-Ibeta, and cAMP was slightly weaker than cGMP in PKG-I activation. VSMC of young rats responded to cAMP or cGMP stimulation in a dose-dependent manner with increasing levels of PKG-I activation. The response was detected within 10 min and continued for at least 24 h. In contrast, VSMC of old rats showed no PKG-I activation during the first hour of cAMP or cGMP stimulation and, at 24 h these cells showed only low-level PKG-I activation. We propose that the reduced PKG-I activation may explain why vascular relaxation is decreased in older individuals.

L3 ANSWER 8 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

TI The biphasic signaling responses of human platelets to cGMP: A predominant role of cAMP-dependent protein kinase in the cGMP-stimulated phosphorylation of **vasodilator-stimulated phosphoprotein**.

AU Li, Zhenyu [Reprint author]; Ajdic, Jasna; Eigenthaler, Martin; Du, Xiaoping

SO Blood, (November 16, 2001) Vol. 98, No. 11 Part 1, pp. 241a. print.

Meeting Info.: 43rd Annual Meeting of the American Society of Hematology, Part 1. Orlando, Florida, USA. December 07-11, 2001. American Society of Hematology.
CODEN: BLOOAW. ISSN: 0006-4971.

AB The roles of cGMP in regulating platelet activation have been controversial since elevation of platelet cGMP levels had been observed in response to either platelet agonists (thrombin, ADP or collagen) or inhibitors (NO or NO donors). Also, while preincubation of cGMP with platelets has been known to inhibit platelet activation via phosphorylation of **vasodilator-stimulated phosphoprotein** (**VASP**), we have recently shown that cGMP promoted platelet glycoprotein Ib-IX (GPIb-IX)-mediated platelet activation by activating the protein kinase G (PKG)-extracellular signal-responsive kinase (ERK) signaling pathway when added together with the agonists without preincubation. To understand the underlying mechanism of the apparent paradoxical roles of cGMP in platelets, we investigated time-dependent platelet response to cGMP by examining the cGMP-induced phosphorylation of two different signaling molecules: ERK and **VASP**. Phosphorylation of ERK was measured by using a polyclonal antibody specifically recognizing the phosphorylated Thr202/Tyr204 site of ERK. Phosphorylation of **VASP** was analyzed by using a **monoclonal** antibody specifically recognizing only phosphorylated Serine239 of **VASP**, 16C2. Here we show that cGMP analogs or nitric oxide (NO) donor glyco-SNAP1 stimulates an early transient ERK phosphorylation at Thr202/Tyr204 that peaks within 1 minute. In contrast, cGMP-induced **VASP** phosphorylation at Ser239 is a delayed but progressive event that reaches maximum only after more than 10 minutes. The cGMP-induced ERK phosphorylation is inhibited by two different inhibitors of PKG (KT5823 or Rp-pCPT-cGMP), suggesting that PKG is responsible for the cGMP-induced ERK phosphorylation. Inhibition of cGMP-induced ERK phosphorylation by KT5823 and Rp-pCPT-cGMP also indicates that these inhibitors are effective in inhibiting PKG function in intact platelets. In contrast to ERK phosphorylation, cGMP-induced phosphorylation at Ser239 of **VASP** is inhibited by protein kinase A (PKA) inhibitors, KT5720, PKI, Rp-8-bromo-cAMP and H89, but not by PKG inhibitors. Similarly, **VASP** (Ser239) phosphorylation induced by NO donor glyco-SNAP1 is also inhibited by PKA inhibitors but not PKG inhibitors. Thus, PKA plays a predominant role in the cGMP-stimulated phosphorylation at Ser239 of **VASP**. Together with our previous studies, these data suggest a novel biphasic signaling mechanism of platelets in response to cGMP: an early PKG-dependent transient activation of ERK pathway that promotes platelet activation, and a delayed cGMP-induced PKA-dependent phosphorylation of **VASP** that are important in cGMP-induced platelet inhibition.

L3 ANSWER 9 OF 15 CAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 5
TI **Monoclonal** antibody against phosphorylated **VASP** (

vasodilator-stimulated phosphoprotein)
IN Eigenthaler, Martin; Hoschutzky, Heinz; Walter, Ulrich
SO PCT Int. Appl., 36 pp.
CODEN: PIXXD2
AB The authors disclose a **monoclonal** antibody (16C2) against **VASP** (**vasodilator-stimulated phosphoprotein**) which only binds **VASP** in its phosphorylated form. In addition, the authors disclose the use of the antibody in the detection of **VASP** in human thrombocytes by flow cytometry and Western blotting. The application of the **monoclonal** antibody, and its fragments, as diagnostic agents and/or therapeutic agents is discussed.

L3 ANSWER 10 OF 15 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI ena/**VASP**-like protein splice variant.
IN CORLEY, N C; GUEGLER, K J; LAL, P
AB US 5912128 A UPAB: 20040218
NOVELTY - An isolated and purified polynucleotide (I) encoding a polypeptide (Ia) comprising 418 amino acids having a regulatory effect on cell trafficking is new.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:
(1) an isolated polynucleotide which is complimentary to (I);
(2) an isolated polynucleotide which encodes ELV1 (a new human ena/**VASP**-like protein splice variant) comprising 1889 base pairs as given in the specification;
(3) an isolated polynucleotide which is complimentary to the polynucleotide of (2);
(4) an expression vector comprising (I);
(5) a host cell comprising the vector of (4);
(6) a method of producing (Ia) which comprises:
(a) culturing the host cell of (5) to express the polypeptide, and
(b) recovering the polypeptide from the host cell culture, and
(7) a method of detecting a polynucleotide encoding (Ia) in a biological sample containing nucleic acids which comprises:
(a) hybridizing the polynucleotide of (1) to at least one of the nucleic acids of the biological sample to form a hybridization complex, and
(b) detecting the complex whose presence correlates with the presence of the polynucleotide encoding the polypeptide in the sample.
USE - ENVL1 is useful in the diagnosis , treatment or prevention of reproductive, immunological, vesicle trafficking, nervous system, developmental, and neoplastic disorders.
Dwg.0/3

L3 ANSWER 11 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 6
TI Analysis and regulation of **vasodilator-stimulated phosphoprotein** serine 239 phosphorylation in vitro and in intact cells using a phosphospecific **monoclonal** antibody.
AU Smolenski, Albert; Bachmann, Christiane; Reinhard, Kathrin; Hoenig-Liedl, Petra; Jarchau, Thomas; Hoschuetzky, Heinz; Walter, Ulrich [Reprint

author]
SO Journal of Biological Chemistry, (Aug. 7, 1998) Vol. 273, No. 32, pp. 20029-20035. print.
CODEN: JBCHA3. ISSN: 0021-9258.
AB The development and functional analysis of a **monoclonal** antibody (16C2) are reported; the antibody recognizes **vasodilator-stimulated phosphoprotein** (**VASP**; an established substrate of both cAMP- and cGMP-dependent protein kinase) only when serine 239 is phosphorylated. **VASP** serine 239 represents one of the best characterized cGMP-dependent protein kinase phosphorylation sites in vitro and in intact cells. Experiments with purified, recombinant human **VASP** and various **VASP** constructs with mutated phosphorylation sites (S157A, S239A, T278A) and experiments with intact cells (human/rat platelets and other cells) treated with cyclic nucleotide-elevating agents demonstrated the specificity of the **monoclonal** antibody 16C2. Quantitative analysis of the **VASP** shift from 46 to 50 kDa (indicating **VASP** serine 157 phosphorylation) and the appearance of **VASP** detected by the 16C2 **monoclonal** antibody (**VASP** serine 239 phosphorylation) in human platelets stimulated by selective protein kinase activators confirmed that serine 239 is the **VASP** phosphorylation site preferred by cGMP-dependent protein kinase in intact cells. Immunofluorescence experiments with human platelets treated with cGMP analogs showed that the 16C2 **monoclonal** antibody also detects **VASP** serine 239 phosphorylation in situ at established intracellular localization sites. Analysis of **VASP** serine 239 phosphorylation by the 16C2 antibody appears to be the best method presently available to measure cGMP-dependent protein kinase activation in intact cells. Also, the 16C2 antibody promises to be an excellent tool for the evaluation of **VASP** function in intact cells.

L3 ANSWER 12 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 7
TI Functional analysis of cGMP-dependent protein kinases I and II as mediators of NO/cGMP effects.
AU Smolenski, Albert; Burkhardt, A. Mick; Eigenthaler, Martin; Butt, Elke; Gambaryan, Stepan; Lohmann, Suzanne M.; Walter, Ulrich [Reprint author]
SO Naunyn-Schmiedeberg's Archives of Pharmacology, (July, 1998) Vol. 358, No. 1, pp. 134-139. print.
CODEN: NSAPCC. ISSN: 0028-1298.
AB NO and cGMP have emerged as important signal transduction mediators of the effects of certain hormones, inter-/intracellular signals, toxins and drugs. However, a major challenge is to define relevant criteria for determining which of the many NO and/or cGMP effects are dependent on cGMP-dependent protein kinases (cGKs). Important criteria include that: (1) the cell types/tissues investigated contain at least one form of cGK which is activated by the cGMP-elevating agent in the intact cell system; (2) specific activators/inhibitors of cGKs mimic/block the effects of cGMP-elevating agents in the intact cell system; and (3) the cGMP effect is absent or blunted in cGK-deficient systems, or can be reconstituted by

the introduction of active cGKs. Previously, analysis of cGK activity in intact cells has been very difficult. However, the analysis of **vasodilator-stimulated phosphoprotein** (**VASP**) phosphorylation by polyclonal antibodies and newly developed **monoclonal** antibodies, each of which specifically recognize different phosphorylation sites, allows the quantitative measurement of cGK activity in intact cells. With the use of these methods, the properties of certain cGK mutants, cGK activators (cGMP, 8-Br-cGMP, 8-pCPT-cGMP) as well as various "specific cGK inhibitors" (KT 5823, Rp-8Br-PET-cGMPS, Rp-8-pCPT-cGMPS, H8 and H89) were investigated. Although these "specific cGK inhibitors" have been widely used to establish or rule out functional roles of cGKs, very few studies have actually addressed the efficiency/specificity of such compounds in intact cells. Our results demonstrate that these inhibitors are useful tools only when used in combination with other experimental approaches and biochemical evidence.

L3 ANSWER 13 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 8

TI Vinculin proteolysis unmasks an ActA homolog for actin-based Shigella motility.

AU Laine, Roney O.; Zeile, William; Kang, Fan; Purich, Daniel L.; Southwick, Frederick S. [Reprint author]

SO Journal of Cell Biology, (1997) Vol. 138, No. 6, pp. 1255-1264. CODEN: JCLBA3. ISSN: 0021-9525.

AB To generate the forces needed for motility, the plasma membranes of nonmuscle cells adopt an activated state that dynamically reorganizes the actin cytoskeleton. By usurping components from focal contacts and the actin cytoskeleton, the intracellular pathogens Shigella flexneri and Listeria monocytogenes use molecular mimicry to create their own actin-based motors. We raised an antibody (designated FS-1) against the FEFPPPPTDE sequence of Listeria ActA, and this antibody: (a) localized at the trailing end of motile intracellular Shigella, (b) inhibited intracellular locomotion upon microinjection of Shigella-infected cells, and (c) cross-reacted with the proteolytically derived 90-kD human vinculin head fragment that contains the Vinc-1 oligoproline sequence, PDFPPPPPDL. Antibody FS-1 reacted only weakly with full-length vinculin, suggesting that the Vinc-1 sequence in full-length vinculin may be masked by its tail region and that this sequence is unmasked by proteolysis. Immunofluoresence staining with a **monoclonal** antibody against the head region of vinculin (Vin 11-5) localized to the back of motile bacteria (an identical staining pattern observed with the anti-ActA FS-1 antibody), indicating that motile bacteria attract a form of vinculin containing an unmasked Vinc-1 oligoproline sequence. Micro-injection of submicromolar concentrations of a synthetic Vinc-1 peptide arrested Shigella intracellular motility, underscoring the functional importance of this sequence. Western blots revealed that Shigella infection induces vinculin proteolysis in PtK2 cells and generates p90 head fragment over the same 1-3 h time frame when intracellular bacteria move within the host cell cytoplasm. We also discovered that microinjected p90, but not full-length vinculin, accelerates rates of pathogen motility by a factor

of 3+-0.4 in Shigella-infected PtK2 cells. These experiments suggest that vinculin p90 is a rate-limiting component in actin-based Shigella motility, and that supplementing cells with p90 stimulates rocket tail growth. Earlier findings demonstrated that vinculin p90 binds to IcsA (Suzuki, T.A., S. Saga, and C. Sasakawa. 1996. J. Biol. Chemical 271:2187821885) and to **vasodilator-stimulated phosphoprotein** (**VASP**) (Brindle, N.P.J., M.R. Hold, J.E. Davies, C.J. Price, and D.R. Critchley. 1996. Biochem. J. 318:753757).

We now offer a working model in which proteolysis unmasks vinculin's ActA-like oligoproline sequence. Unmasking of this site serves as a molecular switch that initiates assembly of an actin-based motility complex containing **VASP** and profilin.

L3 ANSWER 14 OF 15 CAPLUS COPYRIGHT 2004 ACS on STN
TI Antibodies to Listeria monocytogenes
AU Bhunia, Arun K.
SO Critical Reviews in Microbiology (1997), 23(2), 77-107
CODEN: CRVMAC; ISSN: 1040-841X
AB A review with 179 refs. Listeria monocytogenes is one of the leading food-borne pathogens and has been implicated in numerous outbreaks in the last 2 decades. Immunocompromised populations are usually the most susceptible to Listeria infections. Although the pathogenic mechanism is a complex process, significant progress has been made in unraveling the mechanism in recent years. It is now clear that numerous extracellular and cell-associated proteins, such as internalin, listeriolysin, actin polymerization protein, phospholipase, metalloprotease, and possibly p60 proteins, are essential for L. monocytogenes entry into mammalian cells, survival inside the phagosome, escape into the cytoplasm, and cell-to-cell spread. Other proteins may be responsible for growth and physiol. or to maintain the structural integrity of the bacteria. **Monoclonal** and polyclonal antibodies have been developed against many of those antigens or their synthetic derivs. that have helped greatly to determine the structure and function of these antigens. The antibodies were also used for the diagnosis and detection, immunocytochem. staining, and serotyping of Listeria. Humoral immune response to live L. monocytogenes cells was examined in naturally or exptl. infected hosts. Studies revealed that only extracellular antigens induced the humoral response, whereas cell-associated antigens had apparently no response. It is speculated that during the occasional bacteremic phase, L. monocytogenes releases extracellular antigens that are then processed by the immune system for antibody production As L. monocytogenes is an intracellular pathogen, the cell-associated antigens are not persistent in the blood circulation and thus fail to stimulate the humoral immune response.

L3 ANSWER 15 OF 15 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN

10/664,970

TI **Monoclonal** antibodies against the focal adhesion protein **VASP** revealing epitopes involved in the interaction with two **VASP** binding proteins and **VASP** phosphorylation.

AU Abel, Kathrin [Reprint author]; Lingnau, Andreas; Niebuhr, Kirsten; Wehland, Juergen; Walter, Ulrich [Reprint author]

SO European Journal of Cell Biology, (1996) Vol. 69, No. SUPPL. 42, pp. 39. Meeting Info.: 21st Annual Meeting of the German Society for Cell Biology.
Hamburg, Germany. March 24-28, 1996.
CODEN: EJCBDN. ISSN: 0171-9335.

9/10/04